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STUDY PROTOCOL OF A RANDOMIZED CONTROLLED TRIAL TO EVALUATE THE ADD-ON EFFECT OF AYURVEDIC MODULE ON ANTI-INFLAMMATORY CYTOKINE IL-10 IN NIGHT SHIFT WORKERS

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ABSTRACT

Introduction: Different occupations and working conditions produce varied effects on the health of the worker. Nowadays, almost a fifth of the worldwide workforce is engaged in shift work. Shift work, and night shift in particular, is one of the most frequent reasons for the disruption of circadian rhythms, causing significant alterations of sleep and biological functions, which, in turn, can affect the physical and psychological well-being and negatively condition work performance. Night shift work induces sleep deprivation which, in turn, alters the daily levels of alertness and job performance, favoring fatigue. Decreased job satisfaction is strongly associated with increased job stress. Apart from pollution, drugs and pathogens, stress is also found to be a major contributing factor for inflammation. According to past studies, pro-inflammatory cytokines were found to be elevated in highly stressed individuals. Therefore, in the present study an attempt is made to develop and validate the Ayurveda Module for night shift workers and to evaluate the difference in levels of anti-inflammatory marker IL-10 after intervening the Ayurveda module along with Yoga module for lifestyle of night shift workers. **Materials and Methods:** It's a randomized controlled trail comprising two group each of 94 subjects. The participants in the control group will receive the pre-established yoga module as intervention and trail group will receive the newly developed ayurveda module along with yoga module. The primary outcome is to develop and validate the Ayurveda Module and to evaluate the

add on effect of ayurveda module on the levels of anti-inflammatory marker IL-10. The secondary outcome is to evaluate the difference in the levels of PSQI (Pittsburg's Sleep Quality Index) and WHO-QoL (Quality of Life) scores. **Conclusion:** This study can give a good integrative solution for the preventive aspect while dealing with the health issues related with the night shift workers occupation. **Ethics:** Ethical clearance has been taken form the IEC committee, All India Institute of Ayurveda, New Delhi. **Clinical trial registration:** The trail is registered with The Clinical Trail Registry of India (CTRI).

KEYWORDS: Shift disorder, Sleep disease, Healthy lifestyles, Ayurveda Module.

INTRODUCTION

Occupation is an integral part of people's life which has significant impacts on quality of life. People spend about one-third of their lifetime in their working environment.[1] Different occupations and working conditions produce varied effects on the health of the worker. Nowadays, almost a fifth of the worldwide workforce is engaged in shift work.[2] Rotating and scheduling are the main characteristics of shift work. An author has noted that "shift workers are creators and victims at the same time" of this new work organization.[3] Shift work, and night shift in particular, is one of the most frequent reasons for the disruption of circadian rhythms, causing significant alterations of sleep and biological functions, which, in turn, can affect the physical and psychological well-being and negatively condition work performance.[4-6] Many studies have analyzed the impact of shift work on the physical health of workers. Different percentages of shift work disorder have been reported in cross-sectional studies, ranging from 24.4% to 44.3%.[7-9]

Night shift work induces sleep deprivation which, in turn, alters the daily levels of alertness and job performance, favoring fatigue.[10] Moreover, fatigue reduces performance and job satisfaction, favoring absence due to sickness, absenteeism, turnover, and job attrition and often induces use of psychotropic drugs.[11] In a study by Jamal and Baba, rotating shift nurses showed a lower degree of job satisfaction. Decreased job satisfaction is strongly associated with increased job stress.

Apart from pollution, drugs and pathogens, stress is also found to be a major contributing factor for inflammation. According to past studies, pro-inflammatory cytokines were found to be elevated in highly stressed individuals.[12] However, inflammation is body's defense mechanism in response to illnesses, caused due to pathogens. A balance between pro

inflammatory cytokines and anti-inflammatory cytokines is maintained by the body.[13] This balance is crucial for immediate immune response in acute and chronic conditions. Nevertheless, recent studies showed anti-inflammatory effects of exercise. It has been observed, in the past research studies that as anti-inflammatory cytokines increase, pro-inflammatory cytokines decrease with regular exercise. [14-15]

Therefore, interventions that aim to reduce systemic inflammation may not only help in the management of chronic disorders, but also in the prevention of these disorders in individuals having an increased risk.

Sleep is so much important to human beings that it comprises almost 1/3 of one's whole life span. It has a restorative, regenerative, and reparative potential, and hence any compromise to this eventually affects all these functions.[16] Sleep disturbances are debilitating and are being linked to many diseases either as their cause or as manifestations. A global steep rise in noncommunicable diseases is also supposed to be linked with the aberrations in sleep architecture and quality.[16] There are a number of approaches to deal with sleep disturbances including pharmacological and non-pharmacological approaches. Ayurveda, the ancient health-care wisdom of the Orient, gives a deep insight into clinical implications of sleep and also suggests various remedies to cope with sleep disorders. It further proposes many simple home remedies and sleep hygiene propositions which seem promising, are in folklore practice, and have indirect evidences of their effectiveness. Simple remedies like milk, head and foot massage, and *Shirodhara* seem promising in the effective management of a variety of sleep disorders and hence require a thorough review for their possible integration with mainstream medicine.[16]

Study Objectives

The primary objective is to develop and validate the Ayurveda Module for night shift workers and to evaluate the difference in levels of anti-inflammatory marker IL-10 after intervening the Ayurveda module along with Yoga module in the lifestyle of night shift workers. The secondary objective is to is to evaluate the difference in the levels of PSQI (Pittsburg's Sleep Quality Index) and WHO-QoL (Quality of Life) scores after intervening the ayurveda module with yoga module.

MATERIALS AND METHODS

Study Design

The study is a prospective randomized controlled open label clinical trial.

Study Setting

The study participants are being screened and enrolled in the study from the Outpatient department (OPD) of All India Institute of India (AIIA) and field visits too. Training for the yoga module and ayurveda module interventions will be given by the dedicated professionals.

Study duration

The total study duration will be of 3 years including the intervention part of 90 days.

Eligibility criteria

Inclusion criteria

The night shift workers of age between 30 and 45 years irrespective of work profile and gender. He/she should be having night shift duty for more than 07 days per month and should be engaged in this shift duty profession of duration longer than 6 months. Beside this the participant should have PSQI score more than or equal to 5 and should fit to do yoga practices.

Exclusion criteria

Participants with serious neurological and chronic pathological disorders, such as Alzheimer's disease, hypertension, diabetes, and thyroid dysfunctions, as well as pregnant and lactating individuals, will be excluded from the study.

Study Intervention

Study intervention include Yoga module and Ayurveda Module along with Yoga Module in Group A & Group B respectively. Details are as given below.

Group A (Control group)

Participants in this group will undergo a pre-established Yoga Module.[17] Training will be conducted weekly in a face-to-face setting, followed by daily home practice. Participant compliance will be monitored through regular phone calls and the use of a compliance sheet/diary for self-recording. Time of yoga practices will be in the morning during day shift and evening (4 hours after eating) during night shift. Details of Yoga modules are given in the **Table 1**.

Group B (Trial group)

Participants in this group will undergo a pre-established Yoga Module along with Ayurveda Module. A lifestyle module tailored to the unique needs of night shift workers will be developed and validated. This process will encompass three distinct phases: Phase I (Needs Assessment), Phase II (Module Development), and Phase III (Module Validation). For a clearer understanding of the module development and validation process, please refer to **Figure 1**. Training will be given to the research participants at the start of the intervention period and compliance will be assessed through regular phone calls and the use of a compliance sheet/diary for self-recording.

Phase I (Needs Assessment)

This will involve a comprehensive data collection process. A self-administered questionnaire incorporating standardized tools such as the Pittsburgh Sleep Quality Index (PSQI), 24-Hour Dietary Recall, and the Global Physical Activity Questionnaire (GPAQ), along with *Ayurvedic* concepts of *Ahara* (diet), *Nidra* (sleep), and *Dincharya* (daily routine), will be developed. Data will be collected through an online survey using Google Forms, targeting diverse night shift worker populations including healthcare workers, police personnel, defense personnel, corporate employees, and security guards. A consent form will be integrated into the Google Form, ensuring participants provide informed consent before accessing the questionnaire. Contact information will be provided within the consent form for those who require assistance with the questionnaire. To accommodate participants unfamiliar with Google Forms or mobile technology, particularly security personnel, offline data collection using electronic forms will also be implemented.

Phase II (Module Development)

This will involve a comprehensive analysis of the survey data to understand the lifestyle patterns of night shift workers. Based on this analysis, a lifestyle module will be developed, encompassing interventions and general guidelines across key areas. These areas include:

- Sleep: This section will address sleep-related interventions, including optimal sleep timings and sleep hygiene practices.
- Diet: This section will provide comprehensive dietary guidelines, including personalized diet charts.
- Daily Regimens: This section will outline detailed interventions related to daily routines (*Dinacharya*) as per Ayurvedic principles.

- Sleep General Guidelines: This section will provide a list of 'Do's and Don'ts' for improving sleep hygiene and optimizing the sleep environment.
- Diet General Guidelines: This section will provide 'Do's and Don'ts' related to dietary principles, incorporating both Ayurvedic and contemporary scientific perspectives.
- Miscellaneous General Guidelines: This section will address harmful habits and addictions, along with other factors that can significantly impact the health of night shift workers.

Phase III (Module Validation)

This phase will involve a rigorous expert review process. A panel of ten experts with extensive experience in the clinical management of night shift workers will provide feedback on each component of the developed module. A multi-round Delphi method will be employed, with expert ratings on each component scored using a structured format **(Table 2)**. The review process will continue until an overall agreement score of 8 or higher is achieved on each component. Expert selection will be finalized in consultation with the research supervisor, co-supervisor, and research advisors.

Withdrawal criteria: Study participation may be discontinued under several circumstances. These include pregnancy in female subjects, the development of any serious illness necessitating hospitalization and therapeutic intervention, and at the discretion of the principal investigator if they believe it is in the participant's best interest.

Outcome Measures

Primary Outcome measures: The primary outcome measure is to develop and validate the Ayurveda Module for night shift workers and to evaluate the difference in levels of anti-inflammatory marker IL-10 after intervening the Ayurveda module along with Yoga module in the lifestyle of night shift workers.

Secondary Outcome measures: The secondary outcome measure is to evaluate the difference in the levels of PSQI (Pittsburg's Sleep Quality Index) and WHO-QoL (Quality of Life) scores after intervening the ayurveda module along with yoga module.

Safety Outcomes: Regular follow-up visits will be scheduled every 4 weeks apart. Blood samples will be collected with the utmost care. Should any adverse events or side effects occur, immediate attention will be given. Participants will be provided with the study

investigator's contact information for prompt communication. All adverse events will be accurately documented and promptly reported to the ethics committee.

Sample Size Estimation: The sample size was calculated based on study Pratibha Hemant Rajbhoj et. al. 2015. In this study the assumed anti-inflammatory score assessed through IL-10 levels shows that, initial mean score was 0.87 and with a standard deviation of 1.748. The sample size was estimated within 95% confidence interval and 80% power for a two-sided test and with an expected standard deviation of 1.748 from the mean Anti-inflammatory score. In our study, at least an improvement of 1.6 is expected with the intervention. Thus, the estimated sample size is 85 and expecting a 10% dropout, the proposed sample size per group is fixed as 94 for each group. Hence total sample size of the study is 188.

Randomization and allocation

This is an open labelled randomized controlled trial; participants will be assigned randomly to either the control group and trail group in the ration of 1:1 through SNOSE (Sequentially numbered opaque sealed envelope) technique.

Study procedure

This study adheres to a ethical framework to ensure participant safety and data integrity. Prior to trial commencement, all potential participants undergo a comprehensive information session. This session elucidates the study's objectives, procedures, and potential risks in clear, accessible language. To reinforce understanding, written study information and an informed consent form, also presented in easily comprehensible terms, are provided. Only after obtaining voluntary informed consent; participants are assessed for eligibility. This rigorous assessment involves evaluating whether individuals meet predefined inclusion and exclusion criteria. Successful candidates are then randomly assigned to either the control or intervention group. After randomization, participants are formally enrolled in the trial. They are immediately informed of their group assignment and scheduled for baseline assessments. Subsequently, study interventions are dispensed to the groups, accompanied by clear instructions for use and a system for monitoring compliance.

Figure 2 provides a visual overview of the entire study procedure, while **Table 3** details the specific activities conducted during each participant visit.

Laboratory investigations

The biochemical investigation named Interlukin-10 (IL-10) would be done at the screening/ baseline and at the end of the trail i.e. at the end of 12 weeks of intervention.

Data collection, management, and analysis

Data collection will be precisely documented using detailed structured case report forms (CRFs). Each enrolled participant will be assigned a unique identification number, ensuring data traceability across all sources, including CRFs and electronic databases. To safeguard participant confidentiality, consent forms, source documents, and CRFs will be securely stored, while electronic databases will be rigorously protected with robust passwords.

Data entry will employ a double-entry method to enhance accuracy and minimize transcription errors. Access to the collected data will be strictly controlled, limited to the principal investigator, research supervisor, and co-supervisor. To comply with data retention guidelines, source documents and electronic data will be securely archived for a period of five years.

Statistical methods

Data analysis will be conducted using GraphPad Prism. Descriptive statistics, including mean and percentage, will be calculated for all variables. For quantitative data i.e. Laboratory investigation values, between-group comparisons will be assessed using unpaired t-tests, while within-group changes will be evaluated using paired t-tests. For qualitative data i.e. PSQI and WHO-QoL scores, the Mann-Whitney U test will be employed for between-group comparisons, and the Wilcoxon signed-rank test will be used to analyze within-group changes. A two-sided *P* value of <0.05 will be considered statistically significant.

Data Safety and monitoring

To ensure the ethical and scientific integrity of the study, a Departmental research committee (DRC) meeting will arrange every six months. The DRC will rigorously monitor study progress, precisely scrutinize data collection procedures for adherence and appropriateness, and assess the need for any protocol deviations to safeguard the well-being of research participants. Detailed minutes of each DRC meeting will be exactly recorded and archived for future reference and auditability.

Trial drug management and accountability

The study interventions if any comes out through the development and validation process of Ayurveda Module will be stored in a designated storage place and will be labeled, tracked, and dispensed as per the protocol validated. A record would be maintained for the drug dispensed and partially used/unused interventions returned by the participants.

Study compliance

Study compliance will be assessed at each follow-up visit through regular phone calls and the use of a compliance sheet/diary for self-recording from the baseline till the completion of the study. Participants with less than 80% compliance would be withdrawn from the study. Participants will receive consistent follow-up calls to discuss their intervention details, address any concerns, and provide reminders. Personalized counseling sessions will be conducted to educate participants on the importance of adherence, proper medication (if any comes out through Ayurveda module validation process) usage, and managing potential side effects, if any.

Prior and concomitant medication and rescue medication

Participants should generally avoid starting any new medications without first consulting with the researcher, unless it's an emergency. They should also report any unexpected or concerning symptoms to the researcher immediately. While participants can usually continue taking their regular medications, the researcher will maintain a careful record of all medications being taken with reason by participants, including those used for other health conditions.

Table 1: Yoga Module.

	Hold Time	Repetitions/Sets	Focus
Shavasana (Corpse Pose)	2-5 min, increasing 1 min/week	1	Relaxation, rest
Ardha Halasana (Half Plough Pose)	5 sec, increasing 5 sec/week to 45 sec	1	Inversion, backstretch
Viparita Karani (Legs-up-the-Wall Pose)	5 min	1	Inversion, relaxation
Matsyasana (Fish Pose)	15-30 seconds	1	Backbend, chest opener
Setubandhasana (Bridge Pose)	15-30 seconds	3	Backbend, hip opener
Bhujangasana (Cobra Pose)	15-30 seconds	3	Backbend, chest opener
Ardha Shalabhasana	15-30 seconds	3	Backbend, core

(Half Locust Pose)			strengthening
Salabhasana (Locust Pose)	15-30 seconds	3	Backbend, core strengthening
Dhanurasana (Bow Pose)	15-30 seconds	3	Backbend, core strengthening
Vakrasana (Spinal Twist)	30 seconds per side	2-3	Twist, spinal mobility
Gomukhasana (Cow Face Pose)	30 seconds	1	Shoulder opening, hip opener
Paschimottanasana (Seated Forward Bend)	5 sec, increasing 5 sec/week to 45 sec	1	Forward bend, hamstring stretch
Supta Vajrasana (Reclining Thunderbolt Pose)	1-2 minutes	1	Hip opener, digestion
Ustrasana (Camel Pose)	15-30 seconds	1	Backbend, chest opener
Chakrasana (Wheel Pose)	15-30 seconds	1	Backbend, chest opener
Utkatasana (Chair Pose)	30 seconds	3	Strength, balance
Vrikshasana (Tree Pose)	30 seconds per side	3	Balance, focus
Tadasana (Mountain Pose)	30 seconds	3	Alignment, grounding
Anuloma-Viloma (Alternate Nostril Breathing)	5-10 minutes	1 cycle	Breathing, balance
Bhramari (Bee Breath)	10 times	1 set	Relaxation, calming
Ujjayi (Victorious Breath)	11 times	1 set	Breathing, energizing
Kapalbhati (Skull Shining Breath)	5-10 minutes	1 set	Energizing, detoxification
Om Chanting	5 minutes	1 set	Meditation, relaxation

Table 2 Delphi method applied for Expert review.

Evaluation Criteria	Rating (1-10)	Comments
1. General Evaluation		
2. Effectiveness		
3. Components Assessment		
*** Sleep**		
*** Diet**		
*** Daily Regimens (Dinacharya)**		
*** Sleep General Guidelines**		
*** Diet General Guidelines**		
*** Miscellaneous General**		

Guidelines		
4. Feasibility		
5. Suggestions for Improvement		

Table 3 Study schedule.

Components	Screening	Baseline assessment	At the end of 4 weeks	At the end of 8 weeks	At the end of 12 weeks
Informed consent	✓				
PSQI assessment	✓				✓
Demographic and medical history		✓			
Laboratory investigation		✓			✓
Training for Yoga protocol and Ayurveda Module as applicable		✓	✓	✓	
Assessment of ADR			✓	✓	✓
Assessment of study compliance			✓	✓	✓
Rescue medication if needed			✓	✓	✓
Issue of trail drugs of any comes out in module development		✓	✓	✓	

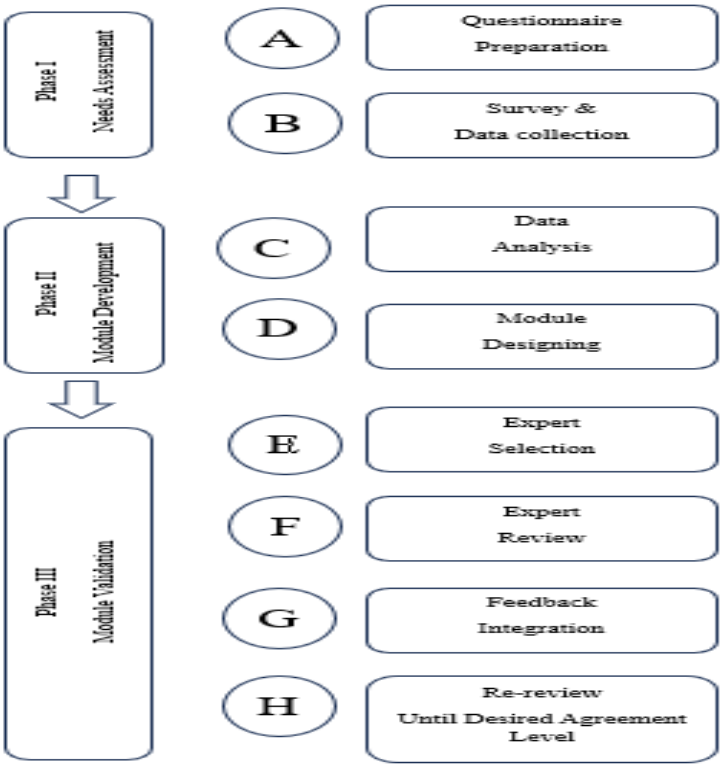

Figure 1 Flow Chart of Module Development and Validation.

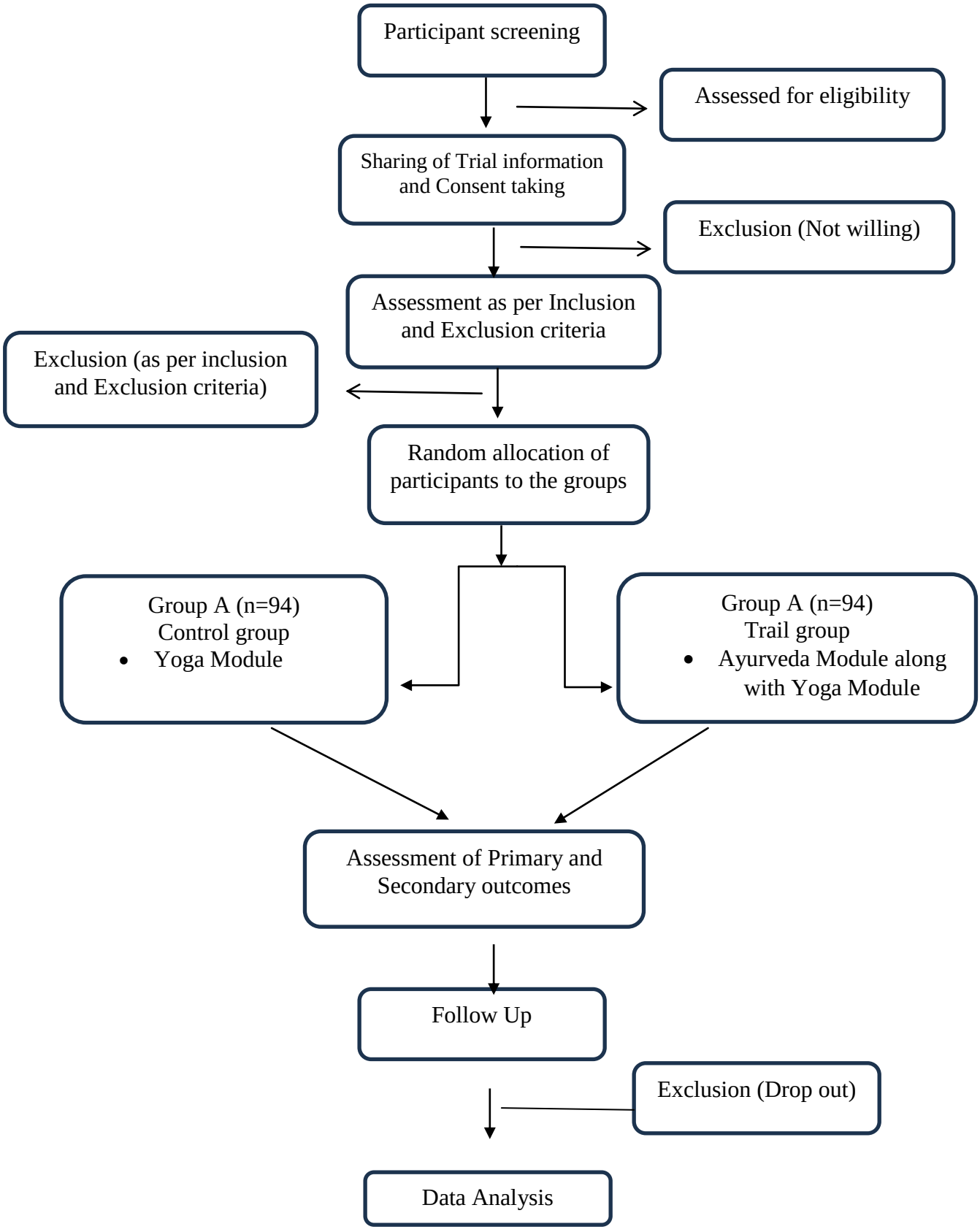

Figure 2 Flow Chart of Study Procedure.

DISCUSSION

In a past study, it was proven that yoga practices could increase anti-inflammatory cytokine in industrial workers. Particularly, industrial workers, who are in direct contact with the pollutants, are affected most due to ill effects of pollutants. Exposure to these pollutants leads to a systemic inflammatory response which can be measured in terms of pro-inflammatory and anti-inflammatory markers.

So, the measures are necessity of the hour that can increase the anti-inflammatory cytokines and decrease the pro-inflammatory cytokines. So, in this present study we will access add on effect of Ayurveda module on one of these markers namely anti-inflammatory cytokine IL-10.

Future prospects

This study has the potential to establish Ayurveda-based modules as effective add-on therapies for improving inflammatory markers like IL-10 in night shift workers. The findings could drive the development of new nutraceuticals, lifestyle protocols, and preventive strategies, raising awareness among healthcare professionals and employers. Ultimately, this study aims to position Ayurveda as a globally recognized complementary approach in occupational health and chronic disease management.

Trial status and dissemination policy

The study is currently in the ongoing phase as of 24/12/2024. The protocol has been approved by the Institutional Review Board (IRB) and Institutional Ethics Committee (IEC) of AIIA, New Delhi. The trial is registered with the Clinical Trials Registry-India (CTRI) under the registration number CTRI/2023/05/052212. All procedures are conducted in accordance with the ethical guidelines outlined by the IRB/IEC and relevant regulatory authorities.

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Ethics committee clearance & no

Ethical clearance has been taken form the IEC committee, All India Institute of Ayurveda,

New Delhi on date 05 January 2023.

Financial support

This study will be completed within sanction/ financial limit of the institute, if more finance is required, the request will be made for due permission and approval of extra budget to the concerned authority AIIA, in the course of research work.

Conflicts of interest: NIL.

AUTHOR CONTRIBUTION

Conceptualization: Dr. Pankaj Kumar

Supervision: Dr. Shivakumar S Harti

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